UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
PREVENTION,
PESTICIDES
AND TOXIC
SUBSTANCES

January 31, 2007

MEMORANDUM

Subject: Efficacy Review for EPA Reg. No. 46781-8, Caviwipes;
DP Barcode: 333784

From: Tajah L. Blackburn, Ph.D., Microbiologist
Efficacy Evaluation Team
Product Science Branch
Antimicrobials Division (7510P)

1/31/07

Thru: Michele Wingfield, Chief
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To: Marshall Swindell PM 33/ Karen Leavy
Regulatory Management Branch I
Antimicrobials Division (7510P)

Applicant: Metrex Research Corporation
28210 Wick Road
Romulus, Michigan 48174

Formulations from Label

Active Ingredient(s)	% by wt.
Diisobutylphenoxyethoxyethyl dimethyl benzyl ammonium chloride.....	0.28%
Isopropanol.....	17.20%
Inert Ingredients.....	82.52%
Total	100.00%

I BACKGROUND

The product, Caviwipes (EPA Reg. No. 46781-8), is a registered towelette with claims of effectiveness against *Mycobacterium bovis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella choleraesuis*, *Trichophyton mentagrophytes*, *Staphylococcus aureus* (MRSA), *Enterococcus faecalis* (VRE), Hepatitis B, Herpes Simplex Virus Type 1 and 2, and Human Immunodeficiency (HIV-1). The registrant requests to make additional claims for effectiveness against *Clostridium difficile* (vegetative cells), *Staphylococcus aureus* (VISA) with reduced susceptibility to vancomycin, Hepatitis C, and Influenza A2 Virus for 2- minute contact time, reduce tuberculocidal contact time from 5 minutes to 3 minutes, and reduce contact time to 2 minutes for previously accepted claims against *Staphylococcus aureus* (MRSA), *Enterococcus faecalis* (VRE), Hepatitis B, Herpes Simplex Virus Type 1 and 2, and Human Immunodeficiency (HIV-1).

The current data packaged contained EPA Form 8570-35, last accepted label (dated July 6, 2006), Statements of No Data Confidentiality for each study, previously submitted studies (MRID Nos. 466309-01, 420358-02, 440227-14, 440227-15, 466309-05, 466309-06, and 466309-09), efficacy studies (MRID Nos. 469453-01 thru -06), and the proposed label.

Previously Submitted Studies for Cavicide (EPA Reg. No. 46781-6)
MRID Nos. 466309-01: AOAC Tuberculocidal Activity of Disinfectants with Cavicide (liquid) (EPA Reg. No. 46781-6)

MRID Nos. 420358-02, Cavicide—Virucidal Efficacy of Micro-Aseptic Products Inc., Cavicide (liquid) Against the Human Immunodeficiency Virus

MRID No. 4402271-14, Cavicide Against Herpes Simplex Virus Type 1, Influenza A virus, Vaccinia virus, Enteroviruses (Polio, Coxsackie B1), and Adenovirus (July 31, 1984)

MRID No. 4402271-15, Cavicide Against Influenza A Virus, Herpes simplex Virus, Vaccinia, Enterovirus, and Adenovirus using Cavicide (July 31, 1984)

MRID No. 466309-05, AOAC Use Dilution Test Supplemental Against *S. aureus* (MRSA) and *E. faecalis* (VRE) using Cavicide (August 18, 2004).

MRID No. 466309-06. Confirmatory Virucidal Effectiveness Using Bovine Diarrhea (BVDV) Surrogate for Human Hepatitis C virus using Cavicide (October 13, 2004)

MRID No. 466309-09. Initial Virucidal Effectiveness Test Using Duck Hepatitis B Virus (DHBV) using Cavicide (September 27, 2004)

II USE DIRECTIONS

This product is designed for disinfecting hard, non-porous surfaces such as high chairs, oxygen hoods, operating tables, spine back boards, slit lamps, tanning beds, hair dryers, hair clippers, shears, razors, brushes, and workstations located in health care setting, bathrooms, daycare centers, emergency vehicles, and salon settings. Directions on the proposed label provided the following information regarding use of the product as a disinfectant and virucide:

Cleaning: Use one towelette to completely preclean surface of all gross debris.

Disinfectant: Use a second towelette to thoroughly wet the surface and allow to remain visibly wet for 3 minutes at room temperature.

Virucide: Use a second towelette to thoroughly wet the surface and allow to remain visibly wet for 2 minutes at room temperature.

III AGENCY STANDARD FOR PROPOSED CLAIMS

Towelette

Towelette products represent a unique combination of antimicrobial chemical and applicator, pre-packaged as a unit in fixed proportions. As such, the complete product, as offered for sale, should be test according to the directions for use to ensure the product's effectiveness in disinfecting hard surfaces. Single-use towelette is intended to be removed from the package, used immediately, and discarded after use. The standard test methods available for hard surface disinfectants (i.e. AOAC Use-Dilution Method, AOAC Germicidal Spray Products as Disinfectants Method), if followed exactly, would not closely simulate the way a towelette product is used. Agency guidelines recommend that a simulated-use test be conducted by modifying the AOAC Germicidal Spray Products as Disinfectants Method. Agency guidelines further recommend that instead of spraying the inoculated surface of the glass slide, the product should be tested by wiping the surface of the glass slide with the saturated towelette, and then subculturing the slides after a specified hold time. Appropriate killing must be demonstrated on all carriers. The above Agency standards are presented in DIS/TSS-1 and EPA Pesticide Assessment Guidelines, Subdivision G, §91-2(h) Pre-saturated or Impregnated towelettes.

Virucides

The effectiveness of virucides against specific viruses must be supported by efficacy data that simulates, to the extent possible in the laboratory, the conditions under which the product is intended to be used. Carrier methods that are modifications or either the AOAC Use-Dilution Method (for liquid disinfectants) or the AOAC Germicidal Spray Products as Disinfectants Method (for spray disinfectants) must be used. To simulate in-use conditions, the specific virus to be treated must be inoculated onto hard surfaces, allowed to dry, and then treated with the product according to the directions for use on the product label. One surface for each of 2 different product lots of disinfectant must be tested against a recoverable virus titer of at least 10^4 from the test surface for a specified exposure period at room temperature. Then, the virus must be assayed by an

appropriate virological technique, using a minimum of four determinations per each dilution assayed. Separate studies are required for each virus. The calculated viral titers must be reported with the test results. For the data to be considered acceptable, results must demonstrate complete inactivation of the virus at all dilutions. When cytotoxicity is evident, at least 3-log reduction in titer must be demonstrated beyond the cytotoxic level. These Agency standards are presented in DIS/TSS-7.

Disinfectants for Use on Hard Surfaces (Additional Bacteria)

Effectiveness of disinfectants against specific bacteria other than those named in the AOAC Use-Dilution Method, AOAC Germicidal Spray Products as Disinfectants Method, AOAC Fungicidal Test, and AOAC Tuberculocidal Activity Method, must be determined by either the AOAC Use-Dilution Method or the AOAC Germicidal Spray Products as Disinfectants Method. Ten carriers must be tested against each specific microorganism with each of 2 product samples, representing 2 different product lots. To support products labeled as "disinfectants" for specific bacteria (other than those bacteria named in the above test method), killing of the specific microorganism on all carriers is required. In addition, plate count data must be submitted for each microorganism to demonstrate that a concentration of at least 10^4 microorganisms survived the carrier-drying step. These Agency standards are presented in DIS/TSS-1.

IV SYNOPSIS OF SUBMITTED EFFICACY STUDIES

1. MRID No. 469453-01, "Testing Pre-Saturated or Impregnated Towelettes for Tuberculocidal Effectiveness" for Caviwipes by Angela Hollingsworth. Study conducted at MicroBioTest. Study completion date—April 21, 2006. Laboratory Project Identification# 198-350.

This study was conducted against *Mycobacterium bovis* (BCG)(Organon Teknika). One lot (Lot No. 5-2258A) of the product Caviwipes was tested using DIS/TSS-6 and EPA Requirements for Pre-Saturated and Impregnated Towelettes for Hard Surface Disinfection. Heat-inactivated horse serum was added to the inoculum to achieve a final concentration of 5%. Carriers were inoculated with test organism, and dried for 20-40 minutes at 37±2°C. Each carrier was wiped right and left for 3 strokes and then up and down for 3 strokes for a total of six strokes per carrier. Between carriers, the area of the towelette used for wiping was rotated to expose a maximum amount of its unused surface. Each towelette set was used to treat ten carriers. Carriers remained wet for 3 minutes. Each carrier was neutralized in 20 ml of MPBM with 7% polysorbate 80 and 1% lecithin, post contact time. Post neutralization, carriers were transferred to 20 ml of MPBM. From each tube, 2 ml were subcultured in Kirchner's medium. All tubes used for primary and secondary transfers were incubated for 60 days at 37±2°C, the results were reported as growth or no growth. If no culture showed visible growth, tubes were incubated for an additional 30 days before final reading. Controls included those for viability, neutralizer effectiveness, sterility, and carrier counts.

2. MRID No. 469453-02, "Testing Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection" for Caviwipes against *Clostridium difficile* (ATCC 9689) by Angela Hollingsworth. Study conducted at MicroBioTest. Study completion date—June 12, 2006. Laboratory Project identification # 198-355.

This study was conducted against *Clostridium difficile* (ATCC 9689). Two lots (Lot Nos. 5-2258A and 5-3257A) of the product, Caviwipes, were tested using the "Germicidal Spray Products as Disinfectants" test as described in the Official Methods of Analysis, Sixteenth edition, 1995, AOAC, DIS/TSS-1, -2, and -3, and the Efficacy Requirements for Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection. Heat-inactivated horse serum was added to the inoculum to achieve a final concentration of 5%. Carriers were inoculated with test organism, and dried for 20-40 minutes at 37±2°C. Each carrier was wiped right and left for 3 strokes and then up and down for 3 strokes for a total of six strokes per carrier. Between carriers, the area of the towelette used for wiping was rotated to expose a maximum amount of its unused surface. Each towelette set was used to treat ten carriers. Carriers remained wet for 2 minutes. Post exposure, carriers were neutralized in 20 ml of RB+. All subculture tubes were incubated for 48±2 hours at 37±2°C. All observations were recorded as growth or no growth. Controls included those for viability, carrier counts, neutralizer effectiveness, sterility, and bacteriostasis.

3. MRID No. 469453-03, "Testing Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection" for Caviwipes against *Staphylococcus aureus* (ATCC 700699) by Angela Hollingsworth. Study conducted at MicroBioTest. Study completion date—April 21, 2006. Laboratory Project identification # 198-358.

This study was conducted against *Staphylococcus aureus* (ATCC 700699) (VISA, reduced susceptibility to vancomycin). Two lots (Lot Nos. 5-2258A and 5-3257A) of the product, Caviwipes, were tested using the "Germicidal Spray Products as Disinfectants" test as described in the Official Methods of Analysis, Sixteenth edition, 1995, AOAC, DIS/TSS-1, -2, and -3, and the Efficacy Requirements for Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection. Heat-inactivated horse serum was added to the inoculum to achieve a final concentration of 5%. Carriers were inoculated with test organism, and dried for 20-40 minutes at 37±2°C. Each carrier was wiped right and left for 3 strokes and then up and down for 3 strokes for a total of six strokes per carrier. Between carriers, the area of the towelette used for wiping was rotated to expose a maximum amount of its unused surface. Each towelette set was used to treat ten carriers. Carriers remained wet for 2 minutes. Post exposure, carriers were neutralized in 20 ml of RB+. All subculture tubes were incubated for 48±2 hours at 37±2°C. All observations were recorded as growth or no growth. Controls included those for viability, carrier counts, neutralizer effectiveness, sterility, bacteriostasis, and antibiotic resistance.

Note: Antibiotic resistance for *S. aureus* (VISA) was attached to the efficacy review.

4. MRID No. 469453-04, "Pre-Saturated or Impregnated Towelette Virucidal Effectiveness Test Using Bovine Viral Diarrhea Virus (BVDV)(Surrogate for Human Hepatitis C Virus)" for Caviwipes, by Lisa Lundberg. Study conducted at MicroBioTest. Study completion date—February 8, 2006. Laboratory Project Identification # 198-354.

This study was conducted against Bovine Viral Diarrhea Virus (BVDV, American BioResearch Laboratories) using MDBK cells (ATCC CCL-22). One lot (Lot No. 5-2258A) of the product, Caviwipes, were tested using the "Germicidal Spray Products as Disinfectants" test as described in the Official Methods of Analysis, Sixteenth edition, 1995, AOAC, DIS/TSS-1, -2, -3, and -7 and the Efficacy Requirements for Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection. An aliquot of 0.1 ml of the viral stock was spread over 1" x 1" square (drying time and temperature requested). Each carrier was wiped right and left for 3 strokes and then up and down for 3 strokes for a total of six strokes per carrier. Between carriers, the area of the towelette used for wiping was rotated to expose a maximum amount of its unused surface. Each towelette set was used to treat ten carriers. Carriers remained wet for 1 minute. After the contact period, the virus-disinfectant was neutralized. This scraped sample was considered to as one $\log_{10}$ dilution. Serial ten-fold dilutions of neutralized virus was prepared in media. Selected dilutions were added to MDBK cells and incubated for to determine the presence or absence of virus. The log reduction of the infectious BVDV FFFU was determined using the Most Probable Number method as described in EPA Statistics Primer (EPA-SP). Controls included those for neutralizer effectiveness, cytotoxicity, cytotoxicity-related viral interference control, plate recovery, viability, and blank towelette control.

5. MRID No. 469453-05, "Pre-Saturated or Impregnated Towelette Virucidal Effectiveness Test" Using Influenza A2 virus for Caviwipes, by Lisa Lundberg. Study conducted at MicroBioTest. Study completion date—January 27, 2006. Laboratory Project Identification # 198-352.

This study was conducted against Influenza A2 virus (SPAFAS) using embryonated chicken eggs (BE Eggs) as the host system. Two lots (Lot Nos. 5-2258A and 5-3257A) of the product, Caviwipes, were tested using the "Germicidal Spray Products as Disinfectants" test as described in the Official Methods of Analysis, Sixteenth edition, 1995, AOAC, DIS/TSS-1, -2, -3, -7 and the Efficacy Requirements for Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection. Viral stock contained ≥5% organic soil. An aliquot of 0.1 ml of the viral stock was spread over 1" x 1" square (drying time and temperature requested). Each carrier was wiped right and left for 3 strokes and then up and down for 3 strokes for a total of six strokes per carrier. Between carriers, the area of the towelette used for wiping was rotated to expose a maximum amount of its unused surface. Each towelette set was used to treat ten carriers. Carriers were remained wet for 30 seconds. After the contact period, the virus-disinfectant was neutralized. This scraped sample was considered to as one $\log_{10}$ dilution. Serial ten-fold dilutions of neutralized virus was prepared in media. Selected dilutions were inoculated in embryonic eggs and incubated for 2-4 days at 37±2°C. The eggs were candled during the incubation period, with allantoic fluid harvesting for hemagglutination assays. Controls included those for neutralizer effectiveness, toxicity, plate recovery, host viability, and blank towelette control.

6. MRID No. 469453-06, "Pre-Saturated or Impregnated Towelette Virucidal Effectiveness Test" Using Duck Hepatitis B Virus for Caviwipes, by Lisa Lundberg. Study conducted at MicroBioTest. Study completion date—March 29, 2006. Laboratory Project Identification # 198-353.

This study was conducted against Duck hepatitis B virus (HepadnaVirus Testing) using primary duck hepatocytes (PDH)(Metzer Farms). One lot (Lot No. 5-2258A) of the product, Caviwipes, were tested using the "Germicidal Spray Products as Disinfectants" test as described in the Official Methods of Analysis, Sixteenth edition, 1995, AOAC, DIS/TSS-1, -2, -3, and -7 and the Efficacy Requirements for Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection. An aliquot of 0.1 ml of the viral stock was spread over 1" x 1" square (drying time and temperature requested). Each carrier was wiped right and left for 3 strokes and then up and down for 3 strokes for a total of six strokes per carrier. Between carriers, the area of the towelette used for wiping was rotated to expose a maximum amount of its unused surface. Each towelette set was used to treat ten carriers. Carriers remained wet for 2 minutes. After the contact period, the virus-disinfectant was neutralized. This scraped sample was considered to as one log10 dilution. Serial ten-fold dilutions of neutralized virus was prepared in media. Selected dilutions were added to PDH and incubated to determine the presence or absence of virus. The log reduction of the infectious DHBV FFFU was determined using the Most Probable Number method as described in EPA Statistics Primer (EPA-SP). Controls included those for neutralizer effectiveness, cytotoxicity, cytotoxicity-related viral interference control, plate recovery, viability, and blank towelette control.

V RESULTS

MRID No. 469453-01. *M. bovis* using Lot. No. 5-2258A

	Results							
Contact time	MPBM +		MPBM		7H9		KM	
	60 days	90 days	60 days	90 days	60 days	90 days	60 days	90 days
3 minutes	0/10	0/10	0/10	0/10	0/10	0/10	0/10	0/10

Carrier count 1.1×10^6 CFU/carrier

MRID No. 469453-02

Test Organisms	Lot No. 5-2258A	Lot No. 5-3257A
C. difficile (vegetative cells)	0/10	0/10

Carrier count 1.1×10^4 CFU/carrier

Test Organisms	Lot No. 5-2258A	Lot No. 5-3257A
S. aureus (VISA)	0/10	0/10

Carrier count 1.8×10^4 CFU/carrier

MRID No. 469453-04

Test Organism	Results			Dried Carrier Count
Bovine Viral Diarrhea Virus		Lot No. 5-2258A (Duplicate 1)	Lot No. 5-2258A (Duplicate 2)	
	10^{-2} and 10^{-7}	Complete inactivation	Complete inactivation	6.52890
	Log MPN/ml	≤1.37985	≤1.37985	

MRID No. 469453-05

Test Organism	Results			Dried Carrier Count
Influenza A2		Lot No. 5-2258A	Lot No. 5-3257A	
	10^{-2} and 10^{-7}	Complete inactivation	Complete inactivation	$10^{7.13}$
	ELD/EID_{50}/ml	≤$10^{1.50}$	≤$10^{1.50}$	

Test Organism	Results			Mean Log_{10}MPN/ml
Duck hepatitis B virus		Lot No. 5-2258A (Duplicate 1)	Lot No. 5-2258A (Duplicate 2)	
	10^{-3} and 10^{-7}	Complete inactivation	Complete inactivation	5.80861
	Log MPN/ml	2.37985	2.37985	

VI CONCLUSIONS

1. The submitted confirmatory efficacy study (469453-01) supports the use of the product Caviwipes as a disinfectant towelette against *M. bovis*, at the ready to use preparation for a contact time of 3 minutes in the presence of organic soil for hard, non-porous surfaces. Initial efficacy study (MRID No. 466309-01) was previously used to support registration. All of the controls met the criteria established for a valid test.

2. The submitted efficacy study (MRID No. 469453-02) supports the use of the product, Caviwipes as a disinfectant towelette against *C. difficile* (vegetative cells) for a contact time of 2 minutes in the presence of organic soil on hard, non-porous surfaces. All controls met the criteria established for a valid test.

3. The submitted efficacy study (469453-03) supports the use of the product, Caviwipes as a disinfectant towelette against *Staphylococcus aureus* (VISA) for a contact time of 2 minutes in the presence of organic soil on hard, non-porous surfaces. All controls met the criteria established for a valid test.

4. The submitted efficacy study (MRID No. 469453-04) supports the use of the product, Caviwipes, as a disinfectant against Bovine Viral Diarrhea Virus (BVDV)(Surrogate for Human Hepatitis C Virus) for a contact time of 1 minute on hard, non-porous surfaces. Initial efficacy study (MRID No. 466309-06) was submitted for the liquid formulation (identical formulation).

5. The submitted efficacy study (MRID No. 469453-05) supports the use of the product, Caviwipes, as a disinfectant against Influenza A2 virus for a contact time of 30 seconds on hard, non-porous surfaces in the presence of organic soil. All controls met the criteria established for a valid test.

6. The submitted efficacy study (MRID No. 469453-06) supports the use of the product, Caviwipes, as a disinfectant with virucidal claims against Duck Hepatitis B Virus (Surrogate for Hepatitis B) for a contact time of 2 minutes on hard, non-porous surfaces in the presence of organic soil. All controls met the criteria established for a valid test.

VII RECOMMENDATIONS

1. The proposed label claims are acceptable regarding the use of Caviwipes as a virucide against Hepatitis C Virus, Influenza A2 Virus, and Hepatitis B virus on hard, non-porous surfaces for a contact time of 2 minutes in the presence of organic soil. Special instructions were extended to Hepatitis B (HBV) and Hepatitis C (HCV) for bloodborne pathogens.

2. The proposed label claims does not include appropriate contact times for the use of Caviwipes as a bactericide against *Staphylococcus aureus* (VISA), *Clostridium difficile* (vegetative), *Staphylococcus aureus* (MRSA), and *Enterococcus faecalis* (VRE). In the registrant's letter (dated September 29, 2005), a request was made to reduce the contact time on the proposed label to 2 minutes. Appropriate studies (previously submitted to support registration) were included, but the label does not reflect this request. Revise accordingly.

3. The proposed label claims does not state appropriate contact times for the use of Caviwipes as a virucide against Influenza A2. In the registrant's letter (dated September 29, 2005), a request was made to add claims of effectiveness for contact time of 2 minutes. Efficacy data was acceptable for a 30 second contact time.

4. Include on the proposed label language requiring the repeated use of the product to ensure that the surface remains wet for the specified contact time.

5. In an Agency letter (December 28, 2005), the following correction was provided "change the statement '...food preparation areas' to read 'non-food contact surfaces in food contact area." Revise accordingly.

6. The proposed label was modified to reduce contact times for ultrasound transducer disinfection from 5 minutes to 3minutes. These claims are acceptable; as supported by the acceptable contact times of ≤ 3minutes for effectiveness against all organisms claimed.

7. Remove the following marketing claims "40% Faster Kill", "Kills TB 40% Faster", and "It's 40% Faster". These quantitative, comparative marketing claims are not acceptable.